Maximal oxygen consumption as related to magnesium, copper, and zinc nutriture[1, 2]

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ABSTRACT Forty-four healthy male university athletes and 20 untrained men underwent maximal treadmill exercise testing to determine the relationship between maximal oxygen consumption and various measurements of elemental nutriture. Hb and mean plasma and erythrocyte concentrations of magnesium, copper, and zinc were within established norms for both groups after a 12-h fast. Mean plasma copper concentration was significantly ($p < 0.01$) higher in the athletes (90 ± 14.3 versus 81 ± 8.0 $\mu g/dl$). Average maximal oxygen consumption also was significantly ($p < 0.001$) higher in the athletes [4.5 ± 0.5 versus 3.3 ± 0.6 L/min and 55.5 ± 7.1 versus 47.0 ± 6.0 ml/(kg·min)]. Plasma magnesium was significantly correlated ($r = 0.46$; $p < 0.002$) with maximal oxygen consumption, ml/(kg·min), among the athletes. This relationship persisted when the effect of Hb concentration was removed by covariance analysis ($p < 0.005$). Only a weak association ($r = -0.32$; $p = 0.17$) was found between oxygen consumption and plasma magnesium in the untrained men. We hypothesize that ionic magnesium may facilitate oxygen delivery to working muscle tissue in trained subjects. *Am J Clin Nutr* 1983;37:407–415.

KEY WORDS Maximal oxygen consumption, magnesium, copper, zinc

Introduction

The role of nutrition in the development and the maintenance of physical work capacity is a continual point of interest in the exercise conscious population. Physical fitness adherents seek advice on nutritional practices from varied sources: peers, coaches, health professionals, and the popular press. Unfortunately, information is limited on the nutritional demands of physical training.

Although the importance of magnesium, copper, and zinc in normal physiological function has been long recognized (1–3), there are few data concerning the relationship of exercise to nutriture of these elements. Several short reports described differences between members of university athletic teams and sedentary students in specific measurements of mineral nutriture (4, 5). The authors concluded that physical training either alters homeostatic mechanisms or produces excessive trace element loss. As these previous reports did not include measurements of physical fitness, we compared indices of magnesium, copper, and zinc nutriture with maximal oxygen consumption, which is an objective index of physical fitness (6) among trained athletes and untrained subjects.

Methods

Forty-four male members of varsity athletic teams at the University of North Dakota participated in this study during the last half of their competitive seasons. The representation of volunteers from various athletic squads was football, eight; basketball, 15; wrestling, five; hockey, four; weight lifting, four; and track and field, eight. Before their participation in this study, these athletes had been involved in at least 1 month of concentrated physical conditioning and 2 months of competition and training. We anticipated that this diverse group of athletes would demonstrate a wide range of maximal oxygen uptake levels indicative of the specificity of each individual sport. Twenty untrained men who were living in the Clinical Research Unit of the Center and participating in other studies were also studied. Usual physical activity habits in these volunteers were assessed by recall during

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an interview. Each man reported occasional participation in some recreational games such as softball, volleyball, bowling, basketball, and golf. Five men had been employed during the previous year in construction or farm work. Five men irregularly jogged 2 to 3 miles/day at an intensity of 9 to 10 min/mile. The other 10 men reported infrequent bouts of stretching exercises, yoga, and calisthenics. All subjects were healthy based on review of medical history, physical examination, and resting 12-lead ECG. After a 12-h overnight fast, the subjects underwent an antecubital venipuncture for determination of routine hematology and measurements of plasma and erythrocyte concentrations of magnesium, copper, and zinc.

The athletes consumed ad libitum diets from foods served in university dining areas. Analyses of their 3-day diet histories indicated the athletes consumed diets balanced in terms of the four major food groups, and none was taking vitamin or mineral supplements. **Table 1** lists the foods consumed by the athletes. Based on this tabulation, the athletes ate conventional diets. The untrained volunteers ate meals designed to meet the Recommended Daily Allowances for all nutrients and prepared in the metabolic kitchen of the Center. The mean ($\pm$ SD) daily energy intakes of the two groups were 4047 ± 874 and 2840 ± 54 kcal/day for the athletes and the volunteers, respectively.

Plasma zinc concentrations were determined directly by flame atomic absorption spectrophotometry after dilution with 2% hydrochloric acid in comparison to standards containing 5% glycerol (7). Other metals were compared with standards containing 2% HCl. Whole blood samples were digested with concentrated nitric and 70% perchloric acids by method (II)A of the Analytical Methods Committee (8). The magnesium, copper, and zinc contents of the digestates were determined by flame atomic absorption spectrophotometry with aqueous calibration standards. The trace metal contents of the erythrocytes were calculated from the whole blood and plasma metal concentrations and hematocrit as follows (7).

$$\text{erythrocyte metal} = \frac{\text{whole blood metal} - \text{plasma metal}\ (1 - \text{hematocrit})}{\text{hematocrit}}$$

After the fasting blood draw, each subject performed a maximal exercise test to exhaustion on the treadmill using the Bruce protocol (9). Heart rate was monitored continuously throughout the test with a bipolar CM-5 ECG lead. Oxygen consumption was measured every minute during the treadmill test with a Beckman Metabolic Measurement Cart (10).[3] The criterion that oxygen consumption showed no further increase or a decrease with increasing workloads was used to establish maximal oxygen uptake levels (6).

All data were analyzed by standard statistical methods (11).

This investigation was approved by the Human Studies Committees of the United States Department of Agriculture and the University of North Dakota, and was conducted in conformity with the Declaration of Helsinki. Each subject gave his written informed consent before participating in this study.

Results

The physical characteristics of the athletes and the untrained volunteers are presented in **Table 2**. The athletes were significantly ($p < 0.001$) younger, taller, and had larger average body mass. The biological importance of this age difference, less than 3 yr, is negligible. Aerobic capacity (VO_2 max) was significantly higher in the athletes whether it was expressed as gross oxygen consumption ($p < 0.001$) or normalized for body weight ($p < 0.001$) than the untrained men. These differences in aerobic capacity emphasize the increased metabolic capacity of the athletes who were trained for maximal performance in contrast to the nonathlete laboratory volunteers who participated in prescribed exercise to maintain their levels of work capacity while living in the Clinical Research Unit.

Table 3 summarizes the fasting Hb and some indices of elemental nutriture of the subjects. All values are within established laboratory norms. The only significant difference between the groups was the higher mean plasma copper concentration ($p < 0.01$) in the athletes. No erythrocyte elemental concentrations are reported for the untrained volunteers because the data were compiled retrospectively from previous studies in which erythrocyte magnesium, copper, and zinc levels were not measured.

Among the athletes, a significant association ($r = +0.31$, $p < 0.05$) was observed between plasma and erythrocyte zinc levels. Also, plasma and erythrocyte copper concentrations were correlated ($r = +0.31$, $p < 0.05$). Plasma and erythrocyte magnesium levels were not significantly related ($r = +0.22$, $p = 0.15$).

No relationship was observed between gross maximal oxygen consumption (L/min) and fasting levels of plasma and erythrocyte magnesium, copper, and zinc among the athletes or the volunteers (**Table 4**). However, a significant association ($r = +0.46$, $p < 0.002$)

[3] Beckman Instruments, Inc, Physiological Measurements Operation, Schiller Park, IL. (Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by the US Department of Agriculture, and does not imply its approval to the exclusion of other products that may also be suitable.)

TABLE 1
Foods consumed by male university athletes

Dairy and egg products
- Buttermilk
- Cheeses (american, blue, cheddar, colby, cottage, cream, mozzarella, parmesan, swiss)
- Cream, sour
- Cream, fluid or whipped
- Cream substitute, nondairy
- Egg
- Ice cream
- Milk (whole, 2% fat, skim, evaporated, chocolate flavored)
- Yogurt (plain and with fruit)

Meat, poultry, and fish
- Bacon
- Beef (roast, steak, ground)
- Chicken
- Fish (cod, haddock)
- Pork (roast, ribs, ham)
- Sausages and luncheon meats (bologna, brown and serve, cervelat, frankfurter, liverwurst, polish, salami, thuringer)
- Shrimp paste, canned
- Turkey

Legumes, nuts, and seeds
- Almonds
- Beans (white, red, canned with pork and tomato sauce)
- Cashew nuts
- Peanuts
- Peanut butter
- Pecans
- Soybeans
- Sunflower seeds
- Walnuts

Grain products
- Biscuits
- Bran flakes with raisins
- Bread (french, raisin, white, whole wheat)
- Cake (chocolate, spice)
- Cookies (brownies, chocolate chip, fig bars, oatmeal, peanut, shortbread, vanilla wafers)
- Corn cereal, ready-to-eat
- Corn chips
- Cornmeal
- Corn tortillas
- Crackers (butter, graham, saltine)
- Doughnuts
- Marcaroni
- Muffins
- Noodles, egg
- Oatmeal
- Oat cereal, ready-to-eat
- Pancakes
- Pies (apple, banana custard, blueberry, cherry, lemon)
- Pretzels
- Rice
- Rice cereal, ready-to-eat
- Rolls and buns

Fruits
- Apples (raw, dried)
- Apple juice
- Apricots
- Bananas
- Cherries, maraschino
- Coconut
- Cranberry juice
- Cranberry sauce
- Dates
- Fruit cocktail, canned
- Grapefruit juice
- Grapes
- Grape juice
- Muskmelons
- Olives
- Oranges
- Orange juice
- Peaches
- Pears
- Pineapple
- Prunes
- Raisins
- Strawberries

Fats and oils
- Butter
- Margarine
- Salad dressings (french, italian, thousand island, mayonnaise)
- Shortening, vegetable
- White sauce

Sugar and sweets
- Cake icings
- Candy (caramels, chocolate, peanut butter, nougat gum drops, hard, marshmallow)
- Chocolate syrup
- Honey
- Jams, preserves and jellies
- Sugar (white, brown)
- Syrup

Mixtures and miscellaneous
- Beer
- Beverage, carbonated
- Beverage, carbonated diet
- Beverage, liquor
- Bouillon
- Carob wheat nut bars
- Chewing gum
- Chili con carne with beans, canned
- Chicken and dumplings in gravy, frozen
- Cinnamon
- Coffee
- Cumin seed
- Garlic
- Gelatin dessert
- Gelatin dessert with fruit
- Lemonade
- Mustard, prepared
- Oregano

TABLE 1 (*continued*)

Rye wafers	Paprika
Spaghetti	Parsley
Wheat flour	Pepper, black
Wheat cereal, ready-to-eat	Pepper, chili sauce
Wheat tortillas	Pizza
Vegetables	Popcorn, butter and salt added
Bean sprouts	Pudding
Beans, green	Rosemary
Broccoli	Salt
Brussels sprouts	Soup, commercial canned (chicken gumbo, chicken noodle, chicken rice, cream of mushroom, tomato, vegetable beef)
Cabbage, red	Spaghetti and meatballs, canned
Carrots	Tapioca cream pudding
Cauliflower	Tartar sauce
Celery	Tea
Coleslaw	Thyme
Corn	Tuna salad
Lettuce	Vinegar
Mushrooms	Wine
Onions	
Peas	
Pepper, sweet, green	
Pickles (dill, sweet)	
Potatoes (baked, boiled, french-fried, scalloped)	
Potato chips	
Potato salad	
Radishes	
Squash, winter	
Tomatoes	
Tomato catsup	
Tomato juice	

TABLE 2
Physical characteristics of trained male athletes and untrained male volunteers

	Athletes	Nonathletes
n	44	20
Age (yr)	20.3 ± 1.5*	23.1 ± 2.8†
Stature (cm)	185.5 ± 13.6	177.2 ± 7.9†
Body mass (kg)	81.8 ± 11.7	71.4 ± 8.3†
Body fat (%)‡	9.3 ± 3.0	10.2 ± 2.4
VO_2 max (L/min)	4.5 ± 0.5	3.3 ± 0.6†
VO_2 max [ml/(kg·min)]	55.5 ± 7.1	47.0 ± 6.0†

* Mean ± SD.
† $p < 0.001$.
‡ Estimated from skinfold thickness according to Durnin and Womersley (12).

TABLE 3
Fasting Hb and concentrations of plasma and erythrocyte magnesium, copper, and zinc in 44 male athletes and 20 untrained men

	Athletes	Nonathletes
Hb (g/dl)	15.4 ± 1.2*	15.7 ± 1.2
Plasma magnesium (mg/dl)	2.0 ± 0.23	2.0 ± 0.15
Plasma copper (μg/dl)	90.0 ± 14.3	81.0 ± 8.0†
Plasma zinc (μg/dl)	87.0 ± 10.0	87.0 ± 8.0
Erythrocyte magnesium (mg/dl)	4.7 ± 0.64	
Erythrocyte copper (μg/dl)	85.0 ± 22.9	
Erythrocyte zinc (μg/dl)	1150.0 ± 152.3	

* Mean ± SD.
† $p < 0.01$.

was present between maximal oxygen uptake expressed per kg body weight and plasma magnesium in the athletes (**Fig 1**). When the effects of Hb concentration were removed by covariance analysis the relationship was still significant ($r = +0.46$, $p < 0.005$). Maximal oxygen consumption, ml/(kg·min), and plasma magnesium were not significantly related in the untrained volunteers (**Fig 2**). When the maximal oxygen consumption and plasma magnesium data of the athletes and the untrained volunteers were combined and then analyzed by multiple regression analysis (13), it was determined that one line was not sufficient to represent the two groups ($F = 15.02$, $p < 0.0001$).

TABLE 4
Correlation coefficients relating maximal oxygen uptake (VO_2 max) to plasma and erythrocyte concentrations of magnesium, copper, and zinc in 44 male athletes and 20 untrained men

	Athletes	Nonathletes
VO_2 max, L/min vs		
Plasma magnesium	+0.22	−0.32
Plasma copper	−0.17	−0.25
Plasma zinc	−0.16	−0.26
Erythrocyte magnesium	−0.16	
Erythrocyte copper	+0.12	
Erythrocyte zinc	−0.16	
VO_2 max, ml/(kg·min) vs		
Plasma magnesium	+0.46*	−0.32
Plasma copper	+0.13	−0.06
Plasma zinc	+0.09	−0.24
Erythrocyte magnesium	−0.01	
Erythrocyte copper	−0.07	
Erythrocyte zinc	−0.05	

* $p < 0.002$.

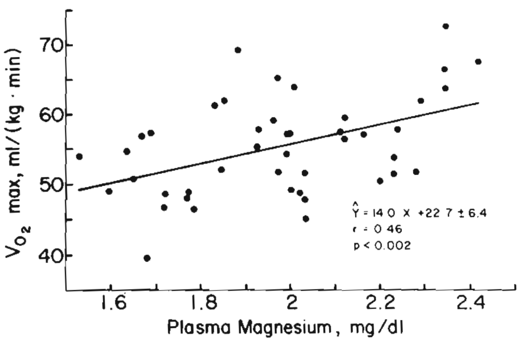

FIG 1. Relationship between maximal oxygen consumption (VO_2 max) and fasting plasma magnesium concentration in 44 male athletes.

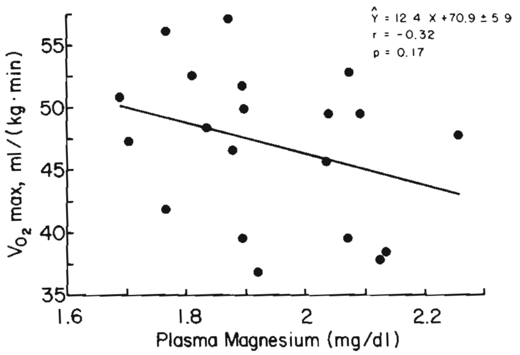

FIG 2. Relationship between maximal oxygen uptake (VO_2 max) and fasting plasma magnesium concentration in 20 untrained men.

Discussion

In this study, the measured indices of mineral status were not lower in the trained male athletes relative to their untrained counterparts. These data conflict with the results of previous studies that have implied that physical training alters elemental nutriture in man. Although no data were provided, Oberleas et al (4) reported in an abstract that zinc and magnesium concentrations in hair and erythrocytes, and magnesium in plasma were lower in collegiate wrestlers than in sedentary students. Semistarvation can affect elemental nutriture adversely. Anderson et al (14) observed reduced heart and skeletal muscle levels of copper and zinc in obese rats that underwent rapid weight loss while consuming a diet high in protein and low in calories. Acute starvation studies in man have demonstrated significant water, nitrogen, and mineral losses, particularly potassium and magnesium, that can impair physical and mental efficiency (15). Wrestlers routinely lose and regain considerable amounts of weight before competition. If this practice of "making weight" is combined with poor eating habits, the potential exists for impaired nutritional status.

Dowdy and Burt (5) followed competitive swimmers through a 6-month training period and observed that average serum ceruloplasmin, a copper-binding protein, fell from 37 IU during the 1st month of training to 26 IU in the 2nd month, and it remained at that level for the remainder of the study. Similarly, serum copper values averaged 64 μg/dl during the first 3 months of the study, and dropped to a mean of 50 μg/dl over the final 3 months of the study.

Surface loss of minerals occurs by a combination of sweat, sebaceous gland secretions, and epidermal exfoliation. Jacob et al (16) studied 13 men living in a controlled environment and reported whole body integumental losses of zinc, copper, and iron of 0.50, 0.30, and 0.33 mg/day which represented 3.9, 26.0, and 2.1% of mean dietary intakes, respectively. Consolazio et al (17) observed significant one-arm zinc and copper sweat losses in men acutely exposed to mild exercise at 37.8 C and 50% relative humidity. Four-day acclimation to these conditions resulted in significant reductions in zinc and copper sweat losses. Sweat losses of magnesium in athletes during endurance exercise have been estimated to be about 1% of the total body content (18). The fall in plasma ceruloplasmin and copper concentrations observed by Dowdy and Burt (5) in swimmers might have resulted from heavy epidermal cell sloughing associated with prolonged water immersion.

Other practices of athletes may influence nutritional status. High carbohydrate, low animal protein diets are popular among endurance athletes seeking to increase muscle glycogen stores and thus improve performance (19). Dressendorfer and Sockolov (20) studied the relationship between training mileage and serum levels of zinc and copper in trained runners. Mean copper concentrations were similar for the runners and the controls (96 and 93 μg/dl, respectively). The runners showed significantly lower average serum zinc concentrations compared to sedentary controls, 76 and 94 μg/dl, respectively. Furthermore, the authors observed that serum zinc was inversely related to training distance. Mean serum zinc levels ranged from 81 to 67 μg/dl for runners training from 6 to 12 and 40 to 84 miles/wk. Eight of the 14 runners who trained at the highest mileage (40 to 84 miles/wk) avoided red meat which is a significant source of dietary zinc. High carbohydrate foods such as pasta, pastry, and fruit are relatively poor sources of dietary zinc (21). Furthermore, zinc obtained from vegetable sources is less available for intestinal absorption than zinc in meat, poultry, or seafood because the presence of phytate and dietary fiber in foods derived from plants (22). Conversely, magnesium is plentiful in green vegetables, and copper is found in organ meats and shellfish. Thus, nutritional practices such as frequent carbohydrate loading or strict vegetarianism may predispose some athletes to mild zinc depletion. The athletes in the present study did not use these methods.

Physical training may induce a redistribution of body minerals. Oh et al (23) showed that plasma zinc levels fell in rats after acute endurance swimming. This decline was related to increased synthesis of hepatic metallothionein, a zinc-binding protein. Dowdy and Dohm (24) reported increased levels of serum copper and ceruloplasmin, a copper-binding protein, after acute exercise in trained and sedentary rats. Furthermore, trained rats had higher mean resting serum ceruloplasmin activities than the corresponding resting controls. Leukocytic endogenous mediator, a hormone-like substance released by phagocytes during exposure to stressors, can lower plasma or serum zinc concentration by causing liver uptake of zinc and stimulate ceruloplasmin release from the liver into the blood (25, 26). Oh et al (23) postulated that reduction of plasma zinc levels in rats after exercise or cold exposure may be mediated by hepatic leukocytic endogenous mediator release. Falchuk (27) has reported effects of ACTH, which has been shown to increase during exercise (28, 29), on liver uptake of zinc that are similar to leukocytic endogenous mediator. Perhaps the significant relationship seen between plasma magnesium and maximal oxygen consumption, ml/(kg·min), in the athletes in this study and the nonsignificant association observed in the untrained volunteers reflects this redistribution.

Any interpretation of data describing physical training and mineral nutritional status is difficult because there are no unequivocal biochemical indices of magnesium, copper, or zinc nutriture. Conflicting data have been presented elsewhere (30) which argue the reliability of plasma and erythrocyte magnesium measurements as indicators of muscle tissue levels during magnesium depletion studies in man and rodents. A similar controversy exists with respect to the interpretation of plasma and erythrocyte copper and zinc levels in mild depletion, in contrast to severe deficiency states (31). However, plasma and erythrocyte measurements of these trace elements are accepted with some reservation as "practical" indices of nutriture.

The elevated plasma copper concentrations of the athletes observed in the present study have been described previously. Haralambie and Keul (32) reported that athletes in training have slightly but significantly higher resting serum copper levels than nonathletes. More recently, Haralambie (33) presented data from 65 trained male athletes and 50 untrained men which confirmed significantly higher ($p < 0.001$) mean serum copper concentrations in the athletes (116 ± 29 versus 93 ± 22 μg/dl). This difference may be related to the role of copper in muscle enzymes, some of which are known to increase in activity following aerobic training (eg, cytochrome oxidase) (34), and a function of copper as ceruloplasmin, as it regulates the transfer of iron from tissues to transferrin (35).

The relationship between plasma magnesium and maximal oxygen consumption in trained athletes observed in the present study suggests the possibility of a metabolic role for magnesium during exercise other than its acknowledged role as a cofactor for many enzymes and in neuromuscular function (1, 30). We propose that magnesium may contribute to the facilitation of oxygen delivery to working muscle by production of 2,3-diphosphoglycerate (2,3-DPG) in the erythrocyte. This contention has support from other experimental data.

Plasma or serum magnesium concentration consistently decreased during and shortly after long duration endurance exercise in marathon runners (36), untrained young men after 2 h of ergometer exercise (33), and well-trained cross-country skiers in conjunction with 90 and 70 km races (37). In the skiers, whole blood magnesium levels were unchanged during the races. Serum magnesium levels declined by 10% after each race with a concomitant increase in erythrocyte magnesium concentration. Both serum and erythrocyte magnesium concentrations were normalized on the days after the races.

In vitro studies have shown correlations between erythrocyte 2,3-DPG and the ATP and the magnesium content of whole blood (38–40). Addition of magnesium ions to anticoagulated whole blood has been shown to increase erythrocyte 2,3-DPG content (41). In hypomagnesemic rats where spherocytic hemolytic anemia was described, decreased erythrocytic magnesium levels and decreased glucose utilization with reduced ATP and 2,3-DPG contents were observed (42).

The mechanism of magnesium action on the regulation of erythrocytic 2,3-DPG levels is not fully understood. In the circulation, when Hb is deoxygenated there is a net increase in the amount of free magnesium ions within the erythrocyte which causes the inhibition of the enzyme hexokinase to be relieved, thereby stimulating glycolysis and further production of 2,3-DPG (41). Brewer (38) has indicated that in vivo only about 5% of the erythrocytic magnesium is unbound, and it is this free magnesium which may regulate glycolysis and 2,3-DPG production.

The correlation coefficient (r = 0.46) calculated for the observed association between maximal oxygen consumption and plasma magnesium in the athletes indicates that 21% of the variance in maximal oxygen uptake was accounted for by the differences in plasma magnesium. Thus other factors account for most of the variance. However, as only 5% of erythrocyte magnesium may regulate 2,3-DPG production, even 21% of the variance may identify an important effect of magnesium.

The observed relationship between maximal oxygen consumption, which is dependent on oxygen delivery to the working muscles, and plasma magnesium in the athletes may represent a cellular adaptation of magnesium metabolism to physical training. Significant increases in erythrocyte 2,3-DPG have been reported in athletes after a single bout of short duration maximal activity (43, 44). Whether these increases in 2,3-DPG were related to magnesium ion redistribution in the blood is not known, but is the objective of future studies.

Our data do not appear to support the belief that physical training per se produces adverse effects on mineral nutriture. Until the hypothesis that chronic physical training does not impair nutritional status has been proven false, it seems reasonable to assume that athletes who consume diets that meet the guidelines of the National Research Council (45) will maintain good nutrition.

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